PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: Robert HOFMEISTER *et al.*	Group Art Unit: 1643
Serial No.: 10/580,660	Examiner: Meera Natarajan
Filed: May 26, 2006	Atty. Dkt. No.: DEBE:066US
For: COMPOSITIONS COMPRISING POLYPEPTIDES	Confirmation No.: 1727

CERTIFICATE OF ELECTRONIC TRANSMISSION
37 C.F.R. § 1.8

I hereby certify that this amendment is being electronically filed with the United States Patent and Trademark Office via EFS-Web on the date below:

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DECLARATION OF THOMAS URBIG UNDER 37 C.F.R. §1.132

Commissioner for Patents
PO Box 1450
Alexandria, VA 22313-1450

I, Dr. Thomas Urbig, do declare that:

1. I am a citizen of Germany residing at Muenchener Str. 27A, D-82131 Gauting . I am the Vice President Process Sciences at Micromet AG, Munich, Germany. A copy of my *curriculum vitae* is attached.

2. Example 6 of Dorken provides that purity of the column fractions has been examined under denaturing conditions. "Purity of column fractions was assessed by ***reducing*** sodium dodecyl sulfate (SDS Bis/Tris 4-12 % polyacrylamide gradient gel electrophoresis (PAGE) employing a MOPS buffer system (Novex)" (emphasis added; page 25, line 51-54). However,

following the teachings of Example 6 of Dorken, multimeric and monomeric bispecific antibody constructs cannot be distinguished, because the multimeric proteins would have all been dissociated into monomeric proteins. Thus, importantly, these methods described in Dorken do not show that Dorken produced a composition comprising the monomer:multimer ratio as recited in claim 26, *i.e.*, a ratio where the multimeric form of the polypeptide constitutes no more than 3% of the total weight of the combined monomeric + multimeric forms of said polypeptide.

3. As shown in Example 3 of the present application, a number of bispecific antibodies were produced in Chinese hamster ovary (CHO) cells according to generally known procedures (Sambrook *et al.*, 1989). Each bispecific single chain antibody produced contains two antigen binding sites, each antigen binding site containing one VH and one VL region. One of the two antigen binding sites in each molecule is specific for the human CD3 antigen. The other antigen binding site (target antigen binding site) is specific for a desired target antigen other than the human CD3 antigen, including *inter alia* CD19. Construct 1 in Table 1 of the present application corresponds to the bispecific construct binding to CD3 and CD19 shown in SEQ ID NO. 1. Ratios of the polypeptide in monomeric/multimeric (here, dimeric) form were determined by a combination of SDS-PAGE performed under reducing conditions, Western Blot performed using Penta-His (Qiagen) and Goat-anti-mouse-AP (Sigma) antibodies and gel filtration performed on a Sephadex S200 column.

4. The relative proportions of bispecific single chain polypeptide present in dimeric form are shown in Table 1 of the present application for polypeptides comprising target antigen specificities against the human CD19 antigen (*i.e.*, the CD3 and CD19 binding construct shown in SEQ ID NO. 1), the human EpCAM antigen, the human Wue1 antigen (a highly specific multiple myeloma antigen) and the human sTn antigen (a carbohydrate displayed on the epithelium of malignant cells in breast, prostate and colon cancers). As can clearly be seen in

Table 1, each bispecific single chain antibody with anti-human CD3 antigen binding specificity, including SEQ ID NO. 1, spontaneously forms significant amounts of multimeric (*i.e.*, here, dimeric) species when left uncontrolled. The propensity to spontaneously form homodimers therefore appears to be a generic characteristic of the class of the bispecific single chain antibodies examined. As such, Dorken's compositions must be presumed to have much higher than 3% concentrations of multimeric proteins.

5. The only way to achieve the compositions of the claimed invention is to follow ***precisely*** the methods of the present invention. The mere use of imidazole gradients, gel filtration, cation exchange chromatography or gel electrophoresis is insufficient to achieve the compositions as now claimed, namely, having "multimeric form of said polypeptide [constituting] no more than 3% of the total weight of the combined monomeric and multimeric forms of said polypeptide."

6. I declare that all statements made herein of my own knowledge are true, and that all statements of my own belief are believed to be true.

July 1st, 2009
Date

Dr. Thomas Urbig

Cited Literature:

[1] Ganapathy V, Smith SB, Prasad PD. Pflügers Arch – Eur J Physiol 447:641-646, 2004.

[2] Rajgopal A, Sierra EE, Zhao R, Goldman ID. Am J Physiol Cell Physiol 281:C1579-C1586, 2001.

[3] Anthony AC. Blood 79:2807-2820, 1992.

[4] Leamon CP, Low PS. Drug Discov Today 6:44-51, 2001.

[5] Sierra EE, Brigle KE, Spinelly MJ, Goldman ID. Biochem Pharamcol 50:1287-1294, 1995.

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Citizenship: Germany
Marital status: married

Curriculum vitae

Professional Experience

Since 2009	Vice President Process Sciences
Seit Februar 2001	Group Leader for Downstream Process Development & Analytical Biochemistry at the Micromet AG (Munich)
1997-2001	Senior Scientist at the Stockholm University, Dept. of Biochemistry, Stockholm, Sweden (Group of Prof. Dr. G. von Heijne)
1995 - 1997	Postdoc at the Harvard University, MCB, Cambridge, MA, USA (Group of Prof. Dr. J. W. Hastings)
1991 - 1994	Ph. D Thesis at the Philipps University, Marburg, Germany (Prof. Dr. M. Marahiel/Prof. Dr. H. Senger)

Educational Experience

1990-1991	Diploma thesis at the Departments of Biochemistry and Biology/Plant Physiology of the Philipps University, Marburg (Prof. Dr. M. Marahiel/Prof. Dr. H. Senger)
1985 - 1990	Studies of Chemistry at the Philipps University, Marburg, Germany

Education

1972-1985	Primary, secondary, and high school

Munich, July 1, 2009